

NANO EXPRESS

Open Access



# LINC01354/microRNA-216b/KRAS Axis Promotes the Occurrence and Metastasis of Endometrial Cancer

Yan Zhang, Wei Zhao, Fei Na, Meng Li and Shengchun Tong\*

## Abstract

**Objective:** LINC01354 has been defined as a tumor driver in several cancers. Nevertheless, whether LINC01354 involves in endometrial cancer (EC) has been little navigated. Thus, the mechanism of LINC01354 was explored in the disease.

**Methods:** Measurements of LINC01354, microRNA (miR)-216b and kirsten rat sarcoma viral oncogene (KRAS) levels in EC tissues and cells were performed. LINC01354 low expression and miR-216b overexpression vectors were introduced into EC cells (Ishikawa), thereby their effects on cell viability, apoptosis, migration and invasion were manifested. Rescue experiments were also carried out by down-regulating LINC01354 and miR-216b spontaneously. Tumorigenesis in vivo was also assessed. The relationships of LINC01354/miR-216b/KRAS were analyzed.

**Results:** Increased LINC01354 and KRAS and reduced miR-216b levels were measured in EC. Silencing LINC01354 or overexpressing miR-216b retarded EC cellular development. LINC01354 counteracted with miR-216b to target KRAS. Suppression of miR-216b antagonized silenced LINC01354-induced impacts on EC cell development. LINC01354/miR-216b/KRAS axis enhanced tumorigenesis in mice with EC.

**Conclusion:** It is testified that silencing LINC01354 inhibits KRAS by up-regulating miR-216b, thereby discouraging cell malignant phenotype in EC.

**Keywords:** Endometrial cancer, LINC01354, MicroRNA-216b, Kirsten rat sarcoma viral oncogene, Tumorigenesis

## Introduction

Endometrial cancer (EC), derived from the epithelium of the uterine cavity, is a prevalent female pelvic malignant tumor, accounting for 4% lifetime incidence [1]. EC patients mainly present well-differentiated cancer in the early stage with favorable prognosis, but aggressive disease subtype remains the great challenge to overcome [2]. Menarche, anovulation, obesity and late menopause can increase estrogen levels and enlarge endometrium, eventually causing endometrial hyperplasia or EC [3]. Surgery (total laparoscopic or laparoscopic hysterectomy and

bilateral salpingo-oophorectomy) is the top 1 treatment for EC [4]. However, for recurrent or metastatic EC, very limited treatment is available [5]. For better control of EC, more effective biomarkers and therapies are urgently required.

Dysregulated long non-coding RNAs (lncRNAs) are believed to connect with tumorigenesis and metastasis in EC [6]. For example, aberrant overexpression of lncRNA RHPN1 antisense RNA 1 [7] and SNHG14 [8] are linked with histological grade and cancer progression in EC. Importantly, lncRNAs-mediated networks are merged as the regulator of EC progression, such as lncRNA RP11-395G23.3-mediated microRNA (miR)-205-5p/phosphatase and tensin homolog axis [9]. LINC01354 is considered as a tumor activator in osteosarcoma, lung

\*Correspondence: Tongshengchun652@163.com  
Department of Gynecology, The Fourth Affiliated Hospital of China Medical University, No. 4, Chongshan East Road, Huanggu District, Shenyang 110032, China

cancer and colorectal cancer (CRC) through promoting cell progression [10–12]. Nevertheless, the defined role of LINC01354, along with its regulatory network has been scarcely ever discussed in EC. miR-216b is the regulated gene by lncRNA that can repress EC cell growth and metastasis [13]. miR-216 has been identified as a key modifier in cervical intraepithelial neoplasia [14] while miR-216-5p in part regulates tumorigenesis of cervical cancer (CC) [15]. Kirsten rat sarcoma viral oncogene (KRAS) is a frequently mutated gene in endometrioid ovarian carcinoma, gastric-type mucinous carcinoma and endometrial mesonephric-like adenocarcinoma [16–18]. KRAS can predict the transition from proliferative endometrium to well-differentiated EC, from further tumor invasion to advanced disease [19]. Actually, KRAS mutation mechanistically mediates the tumorigenesis of EC [20]. KRAS can assess the benignity of precursor or malignant mucinous lesions and distinguish endometrial lesions from cervical lesions [21]. Constructed on the reported researches, this study was initiated to decode the axis of LINC01354/miR-216b/KRAS in EC cell progression.

## Methods and Materials

### Ethics Statement

Our project has been approved by the Ethics Committee of The Fourth Affiliated Hospital of China Medical University. Each patient has issued an informed consent. Procedures and operations performed on animals were consistent with Guidelines for the Care and Use of Laboratory Animals.

### Sample Collection

EC tissues and normal tissues (68 pairs) were collected in The Fourth Affiliated Hospital of China Medical University. Tumor pathology and Federation of Gynecology and Obstetrics stage were analyzed by two pathologists. The samples were frozen in liquid nitrogen and preserved at  $-80^{\circ}\text{C}$  [22].

### Cell Culture

Human EC cell lines (HHUA, KLE, Ishikawa and ECC-1) and normal endometrial (NE) cells acquired from ATCC (VA, USA) were kept in Eagle's Minimum Essential Medium (Gibco, Darmstadt, Germany) consisting of 15% fetal bovine serum (FBS), 100 U/mL penicillin and 100  $\mu\text{g/mL}$  streptomycin [23].

### Cell Transfection

Cells ( $2 \times 10^4$  cells/well) were supposed to culture overnight in a 24-well plate before transient transfection with miR-216b mimic/inhibitor (Applied Biosystems, CA, USA), si-LINC01354 (GenePharma, Shanghai, China),

corresponding negative controls (NC), sh-LINC01354 and miR-216b inhibitor NC, or sh-LINC01354 and miR-216b inhibitor [24].

### Reverse Transcription Quantitative Polymerase Chain Reaction (RT-qPCR)

Total RNA isolated from tissues and cells with Trizol reagent (Life Technologies, Gaithersburg, MD, USA) were quantified by a SmartSpec Plus spectrophotometer (Bio-Rad, Hercules, USA). The target gene was amplified by GoTaq 2-Step RT-qPCR kit (Promega, Madison, USA) and analyzed in Mx3005P qPCR system (Taratagne, CA, USA). Gene expression normalized to GAPDH and U6 was assessed by  $2^{-\Delta\Delta C_t}$  method [23]. Table 1 showed the primer sequences.

### 3-(4, 5-Dimethylthiazol-2-yl)-2, 5-Diphenyltetrazolium Bromide (MTT) Assay

At the 0, 24th, 48th, and 72nd h of culture, respectively, cells ( $1 \times 10^5$  cells/well) in 96-well plates were combined with 15  $\mu\text{L}$  MTT solution (Sigma-Aldrich, MO, USA) to react for 4 h. Then, cells added with dimethyl sulfoxide (200  $\mu\text{L}$ /well) were detected by a microplate reader (Tecan, Maennedorf) to measure optical density<sub>570nm</sub> [25].

### Flow Cytometry

To monitor cell apoptosis, cells were resuspended in  $1 \times$  Binding Buffer (100  $\mu\text{L}$ ), then added with  $1 \times$  Binding Buffer (100  $\mu\text{L}$ ), Annexin V-PE and 7AAD (5  $\mu\text{L}$ ), and incubated in a dark box (BD Bioscience, San Jose, CA, USA). After that, cells supplemented with 250  $\mu\text{L}$  Binding Buffer were detected by a flow cytometer (Fascilibur, BioRad) within 1 h [26].

**Table 1** Primer sequences for genes used in PCR

Genes	Primer sequences
LINC01354	Forward: 5'-GCAATGGTTTGGGCAACTGTAT-3' Reverse: 5'-GAAAAAGCAAGCTGCCATGAGA-3'
miR-216b	Forward: 5'-AAATCTCTGCAGGCAAATGTGA-3' Reverse: 5'-CACCAGGGTCCGAGGT-3'
KRAS	Forward: 5'-GCAATGAGGGACCAAGTACATGAG-3' Reverse: 5'-GTATTGTCCGATCTCCCTACCA-3'
U6	Forward: 5'-GCTTCGGCAGCACATATACTAAAAT-3' Reverse: 5'-CGCTTACGAATTTGCGTGTGAT-3'
GAPDH	Forward: 5'-AACGTGTGAGTGGTGGACCTG-3' Reverse: 5'-AGTGGGTGCTGCTGTTGAAGT-3'

Note: miR-216b, microRNA-216b; KRAS, Kirsten rat sarcoma viral oncogene; GAPDH, glyceraldehyde-3-phosphate dehydrogenase

### Scratch Test

Cells cultured in 6-well plate at  $5 \times 10^5$  cell/mL for 48 h were scratched by a 10- $\mu$ L pipette tip. With removal of the suspended cells, the remaining cells were cultured in serum-free medium for another 48 h, viewed by a MOTOC inverted microscope (Thermo Fisher Scientific, Waltham, USA) and analyzed by IPP (Media Cybernetics, Bethesda, MD, USA) [27].

### Transwell Assay

A transwell incubator (Corning Costar Corp. Corning, USA) was coated with matrix gel, in which cells were cultured ( $3 \times 10^4$  cells/well) in the upper side. A medium containing 20% FBS was set in the lower side. At 24 h post culture, cells that not passed through the filter were removed while those passed were fixed with 4% paraformaldehyde solution (Sigma-Aldrich), stained with 0.01% crystal violet and photographed under a microscope (Olympus BH-2, Tokyo, Japan) [28].

### Dual Luciferase Reporter Gene Assay

Jefferson or Starbase website assessed the binding sites of LINC01354 and miR-216b, and miR-216b and KRAS. In dual luciferase reporter gene assay, LINC01354 or KRAS fragment containing the miR-216b target sequence was inserted into a PmirGLO (GenePharma), thereby LINC01354-WT, LINC01354-MUT, KRAS-WT and KRAS-MUT were obtained. Transfection with the above vector with miR-216b mimic or mimic NC into Lshikawa cells was performed with the help of Lipofectamine 2000 (Invitrogen, USA). Cell luciferase activity was measured by a dual luciferase detection kit (GeneCopoeia, Rockville, USA). Relative luciferase activity = firefly/Renilla luciferase activity [11].

### Western Blot Assay

Extracted from tissues or cells, proteins were quantified by bicinchoninic acid reagent (Beyotime, Shanghai, China). Diluted with  $5 \times$  loading buffer, proteins were denatured at 95 °C, followed by 10% or 12% sodium dodecyl sulphate polyacrylamide gel electrophoresis separation. The proteins were transferred to hybond membranes (Amersham, Munich, Germany) and blocked with 5% skim milk. Anti-KRAS (1:100; ab180772) and anti-GAPDH (1:1000, ab9485, both from Abcam, USA) were the primary antibodies used in protein incubation [29]. After that, the proteins incubated with the corresponding secondary antibody (1:5000; ab205718; Abcam) were visualized by enhanced chemiluminescence reagent (Santa Cruz, CA, USA) [30].

### Tumor Xenografts

BALB/c nude mice (female, 4–5 weeks old) provided by Zhejiang University were injected with the stably-transfected Lshikawa cells ( $1 \times 10^6$  cells/mL, 0.2 mL). The injection was performed at the subscapular area of mice at 8 weeks old. Five mice were utilized in each group. The largest length ( $L$ ) and the width ( $W$ ) perpendicular to the  $L$  were measured every 5 d. Volume =  $0.5 \times L \times W^2$ . All mice were euthanized 30 d later and tumors were weighed and photographed [24].

### Immunohistochemistry

The tumor tissue sections obtained from mice were embedded in paraffin, dissected and baked at 68 °C. Then, the tissues were deparaffinized in conventional xylene, dehydrated with ethanol, and blocked with goat serum. The sections having been incubated with KRAS antibody (1:100; Abcam) were added with diaminobenzidine and hematoxylin successively and observed through a microscope (Nikon, Tokyo, Japan) [31].

### Statistical Analysis

Analyzed SPSS 18.0 software (IBM, NY, USA), the data were presented as mean  $\pm$  standard deviation (repetition = 3). Data calculation utilized t-test or one-way analysis of variance (ANOVA).  $P < 0.05$  was considered statistically significant [27].

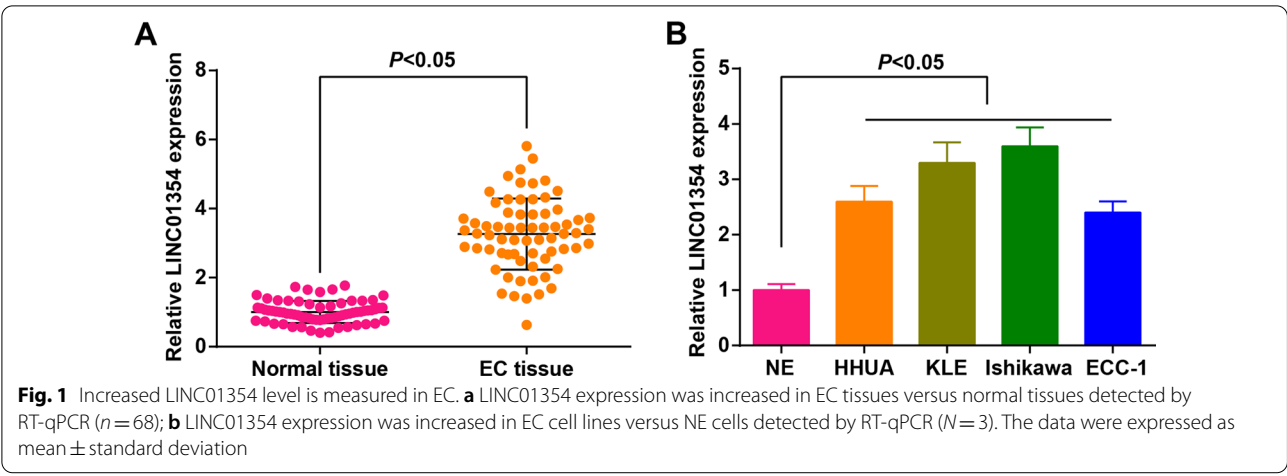
## Results

### Increased LINC01354 Level is Measured in EC

LINC01354 has been revealed to overexpress in non-small cell lung cancer (NSCLC) [11]. As to its role in EC, we firstly measured its level in tissues (normal tissues and EC tissues) and cell lines (normal endometrial cells and EC cell lines Lshikawa, HHUA, KLE and ECC-1). Exactly, LINC01354 level was augmented in EC tissues and cells (Fig. 1a, b). Taking the average of LINC01354 relative expression as the boundary, EC samples were divided into LINC01354 high expression group ( $n = 46$ ) and low expression group ( $n = 22$ ). Clinical analysis demonstrated the connections between LINC01354 expression and tumor differentiation, tumor node metastasis (TNM) stage and lymph node metastasis (LNM) (Table 2).

### Silencing LINC01354 Retards EC Cellular Development

Lshikawa cells with highest LINC01354 expression were applied to cell experiments to further study the role of LINC01354 in EC. RT-qPCR ensured LINC01354 level was inhibited by transfection with sh-LINC01354 in Lshikawa cells (Fig. 2a). Subsequently, LINC01354 down-regulation-induced effects on the



**Fig. 1** Increased LINC01354 level is measured in EC. **a** LINC01354 expression was increased in EC tissues versus normal tissues detected by RT-qPCR ( $n=68$ ); **b** LINC01354 expression was increased in EC cell lines versus NE cells detected by RT-qPCR ( $N=3$ ). The data were expressed as mean  $\pm$  standard deviation

**Table 2** Correlation between the expression of LINC01354 and clinicopathological characteristics of EC

Clinicopathological characteristics	Cases	LINC01354 expression		P
		High (n = 46)	Low (n = 22)	
Age (years)				
< 65	35	23	12	0.7257
≥ 65	33	23	10	
Tumor size (cm)				
< 5	31	24	7	0.1149
≥ 5	37	22	15	
Differentiation				
High	40	21	19	0.0014
Low	28	25	3	
Tumor node metastasis				
I + II	49	29	20	0.0166
III + IV	19	17	2	
Lymph node metastasis				
Yes	45	39	6	< 0.0001
No	23	7	16	

biological processes of Ishikawa cells were studied by MTT assay, flow cytometry, scratch test and Transwell. In Ishikawa cells with knocked down LINC01354, cell viability, invasion and migration were depressed, apoptotic rate was raised (Fig. 2b–e). In summary, silencing LINC01354 can suppress EC cell progression.

#### Reduced miR-216b is Tested in EC; Overexpressing miR-216b Suppresses EC Cell Progression

Next, the downstream targets of LINC01354 involved in EC were explored. miR-216b has been previously discussed to reduce in glioma and breast cancer tissues, and can inhibit cancer cell proliferation, migration and

invasion [32, 33]. In EC, miR-216b level was inhibited in cancer tissues and cell lines (Fig. 3a, b). miR-216b mimic was transfected into Ishikawa cells, after which miR-216b expression was augmented (Fig. 3c). Then, Ishikawa cells with elevated miR-216b expression showed repressed cell progression (Fig. 3d–g). Collectively, miR-216b overexpression limited EC cell malignant phenotype.

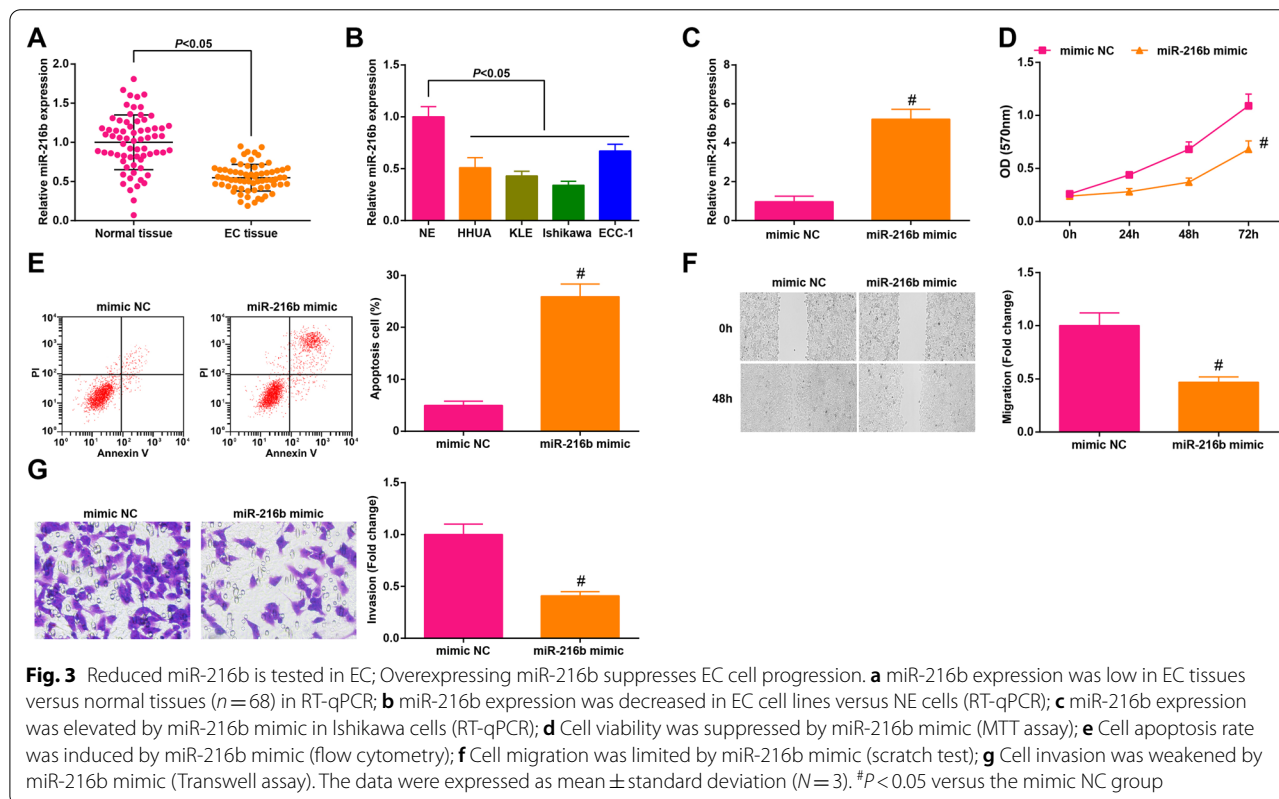
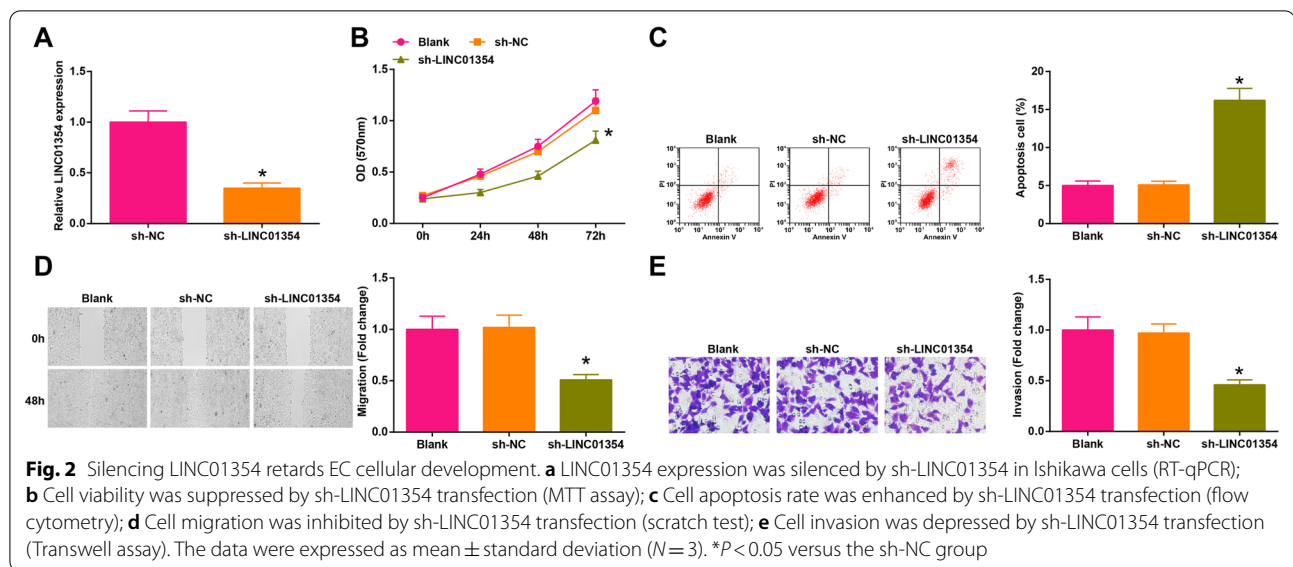
#### LINC01354 Counteracts with miR-216b to Target KRAS

miR-216b is negatively regulated by lncRNAs [34, 35]. RT-qPCR measured an increment in miR-216b expression after down-regulating LINC01354 in cells (Fig. 4a). Therefore, a targeting relation may exist between LINC01354 and miR-216b. Jefferson searched the potential binding sites between LINC01354 and miR-216b (Fig. 4b). Subsequently, dual luciferase report analysis further verified the targeting relationship between LINC01354 and miR-216b, as evident by impaired luciferase activity in cells after co-transfection of LINC01354-WT and miR-216b mimic (Fig. 4c).

Next, miR-216b-mediated downstream genes regulating EC were studied. KRAS expression was enhanced in EC tissues and cell lines (Fig. 4d). After silencing LINC01354 or overexpressing miR-216b in Ishikawa cells, KRAS expression was reduced (Fig. 4e, f). Starbase website showed that miR-216b and KRAS had a targeting relationship (Fig. 4g), and dual luciferase report test manifested that the luciferase activity of KRAS-WT and miR-216b mimic was destructed (Fig. 4h), verifying miR-216b targeting KRAS.

#### Suppression of miR-216b Antagonizes Silenced LINC01354-Induced Impacts on EC Cell Development

The regulation of the LINC01354/miR-216b/KRAS axis on the biological development of EC cells was surveyed. sh-LINC01354 and miR-216b inhibitor

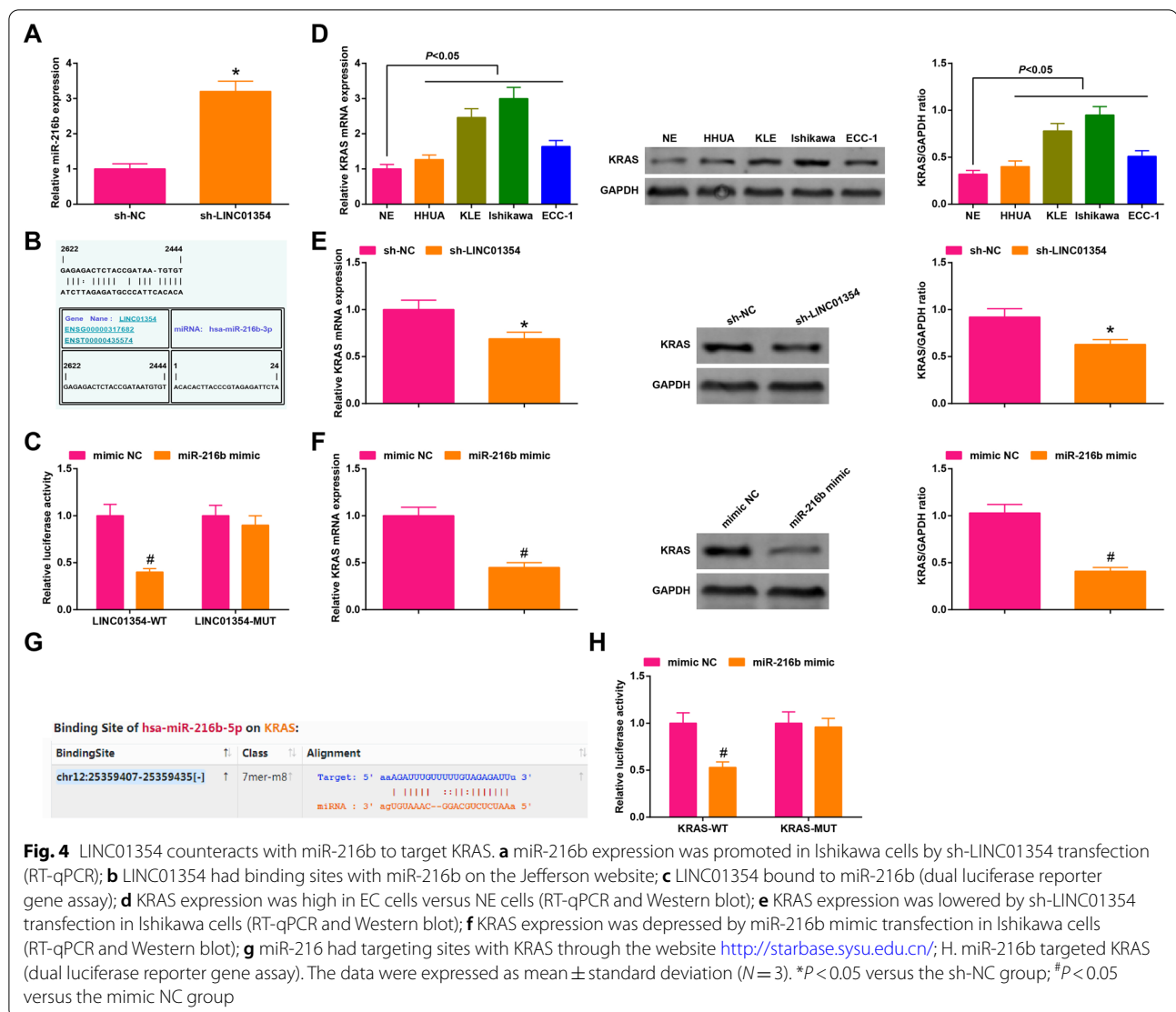


were co-transfected into Ishikawa cells. Then, assays revealed that miR-216b inhibition could negate depleted LINC01354-induced suppression on KRAS expression (Fig. 5a), as well as on Ishikawa cell development (Fig. 5b–e). In brief, LINC01354/miR-216b/KRAS axis regulated EC cell fate.

### Depleting LINC01354 Up-Regulates miR-216b to Slow Down Tumorigenesis in Mice with EC

In vivo growth of EC tumors was observed to further confirm the functional roles of LINC01354 and miR-216b in EC. Ishikawa cells carrying sh-LINC01354 and miR-216b mimic were transplanted into mice and then tumor





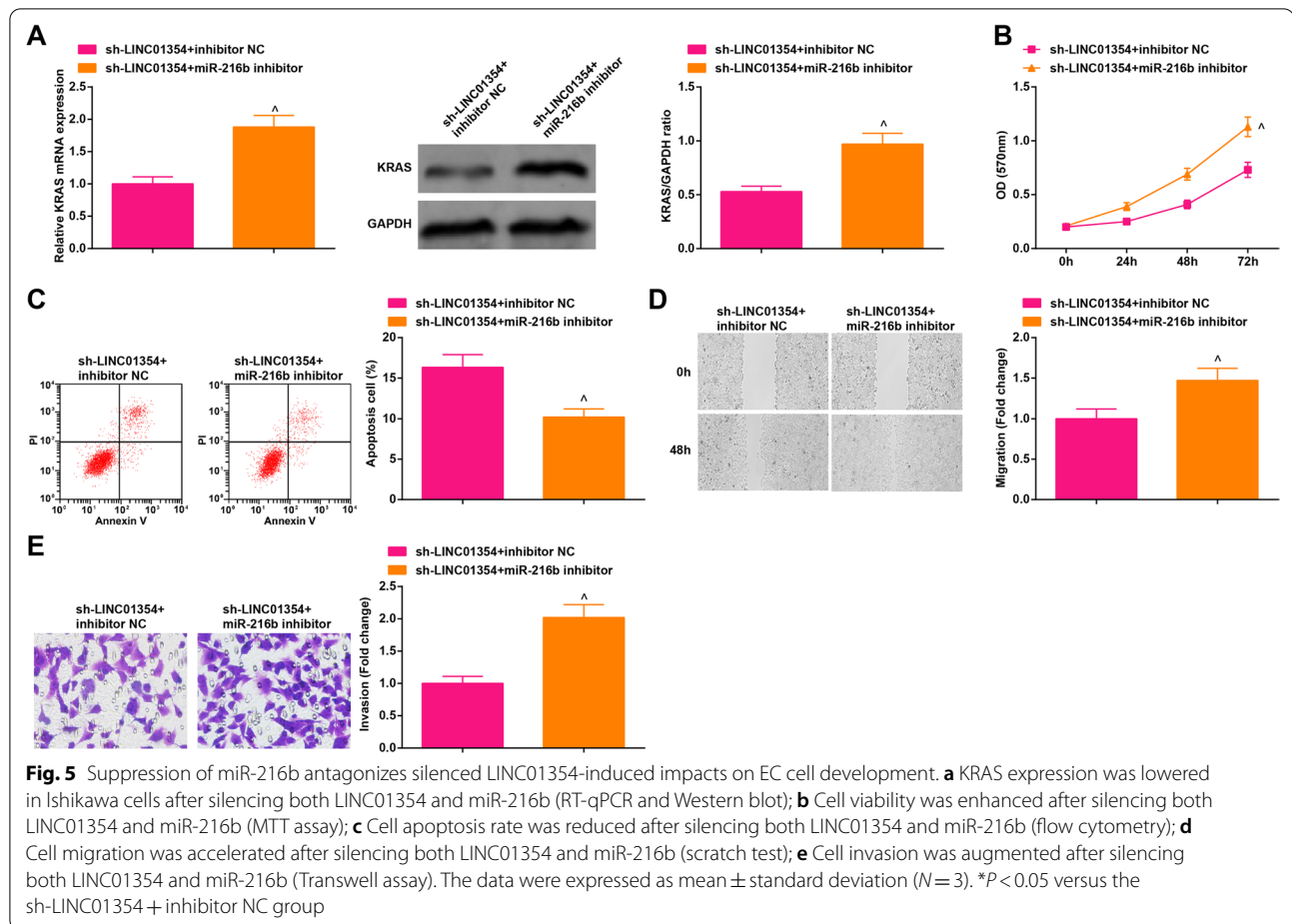
volume and weight were reduced (Fig. 6a–c). In addition, immunohistochemistry demonstrated that silencing LINC01354 or overexpression of miR-216b reduced KRAS expression in EC tumors (Fig. 6d). Shortly, LINC01354/miR-216b/KRAS can regulate the growth of EC tumors in vivo.

## Discussion

EC is the 6th common cancer in female globally whose mortality is largely dependent on tumor recurrence-related poor prognostic factors [36]. As to EC cell progression, this research was pivoted on LINC01354-mediated regulatory network. Firstly, LINC01354 level trended toward up-regulate in EC, which was connected with tumor differentiation, TNM and LNM. Then, silencing LINC01354 in EC cells was proved to be suppressive for cell growth. After that, decreased miR-216b

expression was also investigated in EC and restoring miR-216b limited the acquisition of malignant phenotype of EC cells. Subsequently, inhibiting miR-216b abrogated silenced LINC01354-induced impacts on EC cell development. In a word, LINC01354 suppression elevated miR-216b expression to down-regulate KRAS, thereby restraining cell growth in both vivo and vitro.

At one time, LINC01354 is studied as a competing endogenous RNA in regulating cancer-related pathways in CRC [37]. Incremental LINC01354 level has been once examined in osteosarcoma, and artificially eliminating LINC01354 is conducive for retarding cell invasion in vitro and metastasis in vivo [10]. In the field of NSCLC, the overexpressed LINC01354 is also measured, manifesting a correlation with advanced TNM, while knocking down LINC01354 restricts cancer cells to proliferate and invade [11]. LINC01354 expression goes to an

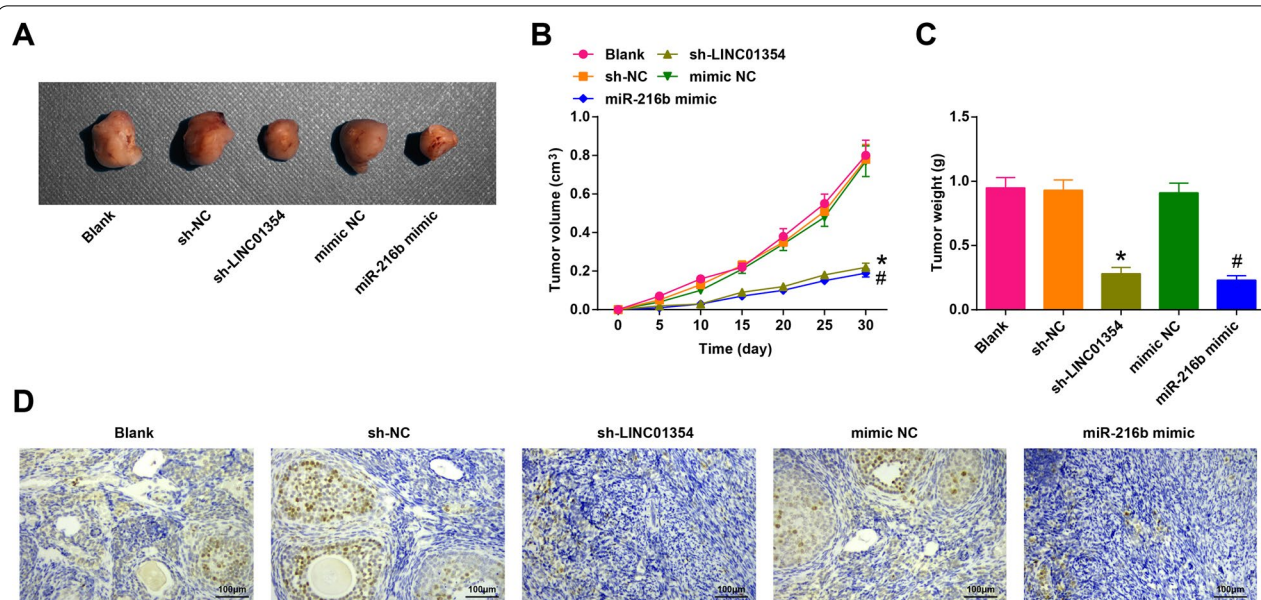


elevation in CRC, and up/down-regulating encourages/discourages cells to form proliferative and migratory phenotypes [38]. As suggested by the aforementioned studies and the present study in combination, LINC01354 indeed is pro-tumor.

Though we identified the binding relation between LINC01354 and miR-216b in the experiments, their reciprocal in diseases needs further confirmation. Regarding to miR-216b, it is implicated to hamper tumors and its under-expression may relate to cancer biology [39]. Of importance, knocking down lncRNA XLOC\_008466 is witnessed to retard proliferation, invasion and migration, as well as drive apoptosis through enhancing miR-216b in CC [40]. Announced in an innovative research, miR-216b expression is reduced and forced expression of miR-216b achieves to destruct cell viability, migration, invasion whereas aggravate apoptosis in EC [13]. Experimentally measured, down-regulated miR-216b showcases in CC while up-regulating miR-216b is the switch for proliferation limitation [41]. LINC00152-mediated miR-216b-5p restoration has been lately confirmed to induce G0/G1 phase cell entry and

apoptosis of CC cells [42]. The regulatory mechanism of miR-216b has not only mentioned in gynecological cancers, but in other cancer types. For instance, restoring the suppressed level of miR-216b in osteosarcoma is promoting for apoptosis induction in vitro [43]. Other than that, in terms of gastric cancer and hepatocellular carcinoma, miR-216b level manifests a reduction in cancer tissues and cells, and forced miR-216b expression induces the restrictions on cancer cell biological activities [44, 45]. In summary, miR-216b itself is the blocker for human cancer development, including but not limited to EC.

KRAS was suggested as the target gene of miR-216b in this EC-focused study, which was supported by reported study findings. Exactly, an inverse correlation exists between miR-216b level and KRAS protein in nasopharyngeal carcinoma, and miR-216b targets KRAS to obstruct cell aggressiveness and tumor formation [46]. Further proved currently, miR-216b-targeted KRAS down-regulation is the inhibitor for pancreatic cancer cell progression [47, 48]. Notably, clear cell renal cell carcinoma cell proliferation and invasion, as well as tumor growth suppression are ascribed to KRAS



**Fig. 6** Depleting LINC01354 up-regulates miR-216b to slow down tumorigenesis in mice with EC. **a** Tumor photos obtained after silencing LINC01354 or overexpressing miR-216b; **b** Tumor volume was decreased after silencing LINC01354 or overexpressing miR-216b; **c** Tumor weight was suppressed after silencing LINC01354 or overexpressing miR-216b; **d** KRAS expression was reduced after silencing LINC01354 or overexpressing miR-216b (IHC). The data were expressed as mean  $\pm$  standard deviation ( $n = 5$ ). \* $P < 0.05$  versus the sh-NC group; # $P < 0.05$  versus the mimic NC group

down-regulation induced by miR-216b [49]. Recurrent KRAS mutation is tested in mesonephric adenocarcinoma [50], and KRAS amplification and its mRNA expression are measured in early stage of recurrent endometrioid EC [51]. In tanshinone I-treated CC cells, KRAS overexpression can enhance cell proliferation [52]. Anyway, miR-216b-mediated KRAS has been implied to manage cancer development and silencing of KRAS restrains tumorigenesis.

## Conclusion

To conclude, the present study makes it comprehensive that LINC01354 raises KRAS expression through binding to miR-216b, thereafter stimulating tumorigenic aggravation in EC. Supplemented by the present study, the mechanism of lncRNA-mediated networks in EC has been further understood. Studies are at wanting in larger scales to further develop the results obtained.

## Abbreviations

EC: Endometrial cancer; miR: MicroRNA; KRAS: Kirsten rat sarcoma viral oncogene; CRC: Colorectal cancer; CC: Cervical cancer; KRAS: Kirsten rat sarcoma viral oncogene; FBS: Fetal bovine serum; RT-qPCR: Reverse transcription quantitative polymerase chain reaction; NSCLC: Non-small cell lung cancer; TNM: Tumor node metastasis.

## Authors' Contributions

ST finished study design, YZ, WZ, FN finished experimental studies, YZ, ML finished data analysis, YZ finished manuscript editing. All authors read and approved the final manuscript.

## Funding

No funds, grants, or other support was received.

## Availability of Data and Materials

The original contributions presented in the study are included in the article/Supplementary Material, further inquiries can be directed to the corresponding author.

## Declarations

### Ethics Approval and Consent to Participate

This study was approved and supervised by the animal ethics committee of The Fourth Affiliated Hospital of China Medical University. The treatment of animals in all experiments conforms to the ethical standards of experimental animals.

### Consent for Publication

Patients signed informed consent regarding publishing their data and photographs.

### Competing interests

The authors have no conflicts of interest to declare that are relevant to the content of this article.

Received: 2 March 2021 Accepted: 15 December 2021

Published online: 31 January 2022



## References

- Tempfer CB et al (2020) Menopausal hormone therapy and risk of endometrial cancer: a systematic review. *Cancers (Basel)* 12(8):2195
- Dork T et al (2020) Genetic susceptibility to endometrial cancer: risk factors and clinical management. *Cancers (Basel)* 12(9):2407
- Francies FZ et al (2020) Genomics and splicing events of type II endometrial cancers in the black population: racial disparity, socioeconomic and geographical differences. *Am J Cancer Res* 10(10):3061–3082
- van den Heerik A, et al (2021) Adjuvant therapy for endometrial cancer in the era of molecular classification: radiotherapy, chemoradiation and novel targets for therapy. *Int J Gynecol Cancer* 31(4):594–604.
- Megino-Luque C et al (2020) Small-molecule inhibitors (SMIs) as an effective therapeutic strategy for endometrial cancer. *Cancers (Basel)* 12(10):2751
- Liu H, Wan J, Chu J (2019) Long non-coding RNAs and endometrial cancer. *Biomed Pharmacother* 119:109396
- Zhang XJ, et al (2021) lncRNA RHPN1-AS1 promotes the progression of endometrial cancer through the activation of ERK/MAPK pathway. *J Obstet Gynaecol Res* 47(2):533–543
- Wang GF, Wen LN (2020) lncRNA SNHG14 promotes proliferation of endometrial cancer through regulating microRNA-655-3p. *Eur Rev Med Pharmacol Sci* 24(20):10410–10418
- Xin W et al (2020) lncRNA RP11–395G23.3 suppresses the endometrial cancer progression via regulating microRNA-205–5p/PTEN axis. *Am J Transl Res* 12(8):4422–4433
- Zhou C et al (2020) COX10-AS1 facilitates cell proliferation and inhibits cell apoptosis in glioblastoma cells at post-transcription level. *Neurochem Res* 45(9):2196–2203
- Yang G et al (2019) LINC01354 enhances the proliferation and invasion of lung cancer cells by regulating miR-340-5p/ATF1 signaling pathway. *Artif Cells Nanomed Biotechnol* 47(1):3737–3744
- Wang Z et al (2019) AKT drives SOX2 overexpression and cancer cell stemness in esophageal cancer by protecting SOX2 from UBR5-mediated degradation. *Oncogene* 38(26):5250–5264
- Xie P et al (2017) Knockdown of lncRNA CCAT2 inhibits endometrial cancer cells growth and metastasis via sponging miR-216b. *Cancer Biomark* 21(1):123–133
- Yang C, Xu X, Jin H (2016) Identification of potential miRNAs and candidate genes of cervical intraepithelial neoplasia by bioinformatic analysis. *Eur J Gynaecol Oncol* 37(4):469–473
- Zhu H et al (2018) SNHG16/miR-216-5p/ZEB1 signal pathway contributes to the tumorigenesis of cervical cancer cells. *Arch Biochem Biophys* 637:1–8
- Hollis RL et al (2020) Molecular stratification of endometrioid ovarian carcinoma predicts clinical outcome. *Nat Commun* 11(1):4995
- Park E et al (2020) Genetic characteristics of gastric-type mucinous carcinoma of the uterine cervix. *Mod Pathol* 34:637–646
- Horn LC et al (2020) Mesonephric-like adenocarcinomas of the uterine corpus: report of a case series and review of the literature indicating poor prognosis for this subtype of endometrial adenocarcinoma. *J Cancer Res Clin Oncol* 146(4):971–983
- Sideris M et al (2019) The role of KRAS in endometrial cancer: a mini-review. *Anticancer Res* 39(2):533–539
- Xiong J et al (2016) The clinical significance of K-ras mutation in endometrial “surface epithelial changes” and their associated endometrial adenocarcinoma. *Gynecol Oncol* 142(1):163–168
- He M et al (2015) KRAS mutations in mucinous lesions of the uterus. *Am J Clin Pathol* 143(6):778–784
- Du Y et al (2018) lncRNA DLEU1 contributes to tumorigenesis and development of endometrial carcinoma by targeting mTOR. *Mol Carcinog* 57(9):1191–1200
- Ly Y et al (2019) Upregulation of long non-coding RNA OGFRP1 facilitates endometrial cancer by regulating miR-124-3p/SIRT1 axis and by activating PI3K/AKT/GSK-3beta pathway. *Artif Cells Nanomed Biotechnol* 47(1):2083–2090
- Zhou YX et al (2018) Long noncoding RNA HOTAIR mediates the estrogen-induced metastasis of endometrial cancer cells via the miR-646/NPM1 axis. *Am J Physiol Cell Physiol* 314(6):C690–C701
- Fang Q, Sang L, Du S (2018) Long noncoding RNA LINC00261 regulates endometrial carcinoma progression by modulating miRNA/FOXO1 expression. *Cell Biochem Funct* 36(6):323–330
- Sun KX et al (2017) lncRNA MEG3 inhibit endometrial carcinoma tumorigenesis and progression through PI3K pathway. *Apoptosis* 22(12):1543–1552
- Hu S et al (2019) Overexpression of lncRNA PTENP1 suppresses glioma cell proliferation and metastasis in vitro. *Onco Targets Ther* 12:147–156
- Wang Y et al (2019) The lncRNA UNC5B-AS1 promotes proliferation, migration, and invasion in papillary thyroid cancer cell lines. *Hum Cell* 32(3):334–342
- Wang YG et al (2018) lncRNA DGCR5 represses the development of hepatocellular carcinoma by targeting the miR-346/KLF14 axis. *J Cell Physiol* 234(1):572–580
- Chen S et al (2018) lncRNA TDRG1 enhances tumorigenicity in endometrial carcinoma by binding and targeting VEGF-A protein. *Biochim Biophys Acta Mol Basis Dis* 1864(9 Pt B):3013–3021
- Chen HX et al (2017) MicroRNA-29b inhibits angiogenesis by targeting VEGFA through the MAPK/ERK and PI3K/Akt signaling pathways in endometrial carcinoma. *Cell Physiol Biochem* 41(3):933–946
- Zhang T et al (2017) miR-216b inhibits glioma cell migration and invasion through suppression of FoxM1. *Oncol Rep* 38(3):1751–1759
- Menbari MN et al (2019) MiR-216b-5p inhibits cell proliferation in human breast cancer by down-regulating HDAC8 expression. *Life Sci* 237:116945
- Liu F et al (2019) lncRNA DSCAM-AS1 downregulates miR-216b to promote the migration and invasion of colorectal adenocarcinoma cells. *Onco Targets Ther* 12:6789–6795
- Sun S et al (2020) Long noncoding RNA LINC00265 promotes glycolysis and lactate production of colorectal cancer through regulating of miR-216b-5p/TRIM44 axis. *Digestion* 101(4):391–400
- Coll-de la Rubia E et al (2020) Prognostic biomarkers in endometrial cancer: a systematic review and meta-analysis. *J Clin Med* 9(6):1900
- Wang X et al (2018) A 15-lncRNA signature predicts survival and functions as a ceRNA in patients with colorectal cancer. *Cancer Manag Res* 10:5799–5806
- Li J et al (2019) LINC01354 interacting with hnRNP-D contributes to the proliferation and metastasis in colorectal cancer through activating Wnt/beta-catenin signaling pathway. *J Exp Clin Cancer Res* 38(1):161
- Jana S et al (2020) Therapeutic targeting of miRNA-216b in cancer. *Cancer Lett* 484:16–28
- Guo F et al (2018) Long non-coding RNA XLOC\_008466 acts as an oncogenic molecular in cervical cancer tumorigenesis. *Biomed Pharmacother* 98:88–94
- He S et al (2017) MiR-216b inhibits cell proliferation by targeting FOXM1 in cervical cancer cells and is associated with better prognosis. *BMC Cancer* 17(1):673
- Zheng JJ et al (2019) Long non-coding RNA 00152 promotes cell proliferation in cervical cancer via regulating miR-216b-5p/HOXA1 axis. *Eur Rev Med Pharmacol Sci* 23(9):3654–3663
- Yang D et al (2020) microRNA-216b enhances cisplatin-induced apoptosis in osteosarcoma MG63 and SaOs-2 cells by binding to JMJD2C and regulating the HIF1alpha/HES1 signaling axis. *J Exp Clin Cancer Res* 39(1):201
- Chen X et al (2020) MicroRNA-216b regulates cell proliferation, invasion and cycle progression via interaction with cyclin T2 in gastric cancer. *Anticancer Drugs* 31(6):623–631
- Zhang JF (2020) MicroRNA-216b suppresses the cell growth of hepatocellular carcinoma by inhibiting Ubiquitin-specific peptidase 28 expression. *Kaohsiung J Med Sci* 36(6):423–428
- Deng M et al (2011) miR-216b suppresses tumor growth and invasion by targeting KRAS in nasopharyngeal carcinoma. *J Cell Sci* 124(Pt 17):2997–3005
- Wu X et al (2018) MiR-216b inhibits pancreatic cancer cell progression and promotes apoptosis by down-regulating KRAS. *Arch Med Sci* 14(6):1321–1332
- Ferino A et al (2018) MicroRNA therapeutics: design of single-stranded miR-216b mimics to target KRAS in pancreatic cancer cells. *RNA Biol* 15(10):1273–1285
- Wang Y et al (2018) miR-216b Post-transcriptionally downregulates oncogene KRAS and inhibits cell proliferation and invasion in clear cell renal cell carcinoma. *Cell Physiol Biochem* 49(5):1755–1765
- Mirkovic J et al (2018) Targeted genomic profiling reveals recurrent KRAS mutations in mesonephric-like adenocarcinomas of the female genital tract. *Am J Surg Pathol* 42(2):227–233

51. Iavazzo C, Gkegkes ID, Vrachnis N (2014) Early recurrence of early stage endometrioid endometrial carcinoma: possible etiologic pathways and management options. *Maturitas* 78(3):155–159
52. Dun S, Gao L (2019) Tanshinone I attenuates proliferation and chemoresistance of cervical cancer in a KRAS-dependent manner. *J Biochem Mol Toxicol* 33(4):e22267

### Publisher's Note

Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

**Submit your manuscript to a SpringerOpen<sup>®</sup> journal and benefit from:**

- Convenient online submission
- Rigorous peer review
- Open access: articles freely available online
- High visibility within the field
- Retaining the copyright to your article

---

Submit your next manuscript at ► [springeropen.com](https://www.springeropen.com)

---